

Sub-Task 5

Sampling Procedures for MEASURING DISSOLVED OXYGEN PROFILES

General Guidelines

- **Dissolved oxygen (DO) will be measured at four or more depths in the lake. The near-surface sample can be collected by hand. The other samples require the use of a water sampler.**
- **Depending on the equipment you have to measure DO you will either need a chemical kit or a DO probe (if using a DO probe, follow the procedures for “Measuring Dissolved Oxygen Profiles with a DO Meter” located at the end of the chemical kit procedures).**
- **The chemical kit employs a titration method. Titration is simply dripping a precisely measured amount of chemical into a solution until you achieve a chemical reaction. Because the titration requires you to be very precise, you will test a duplicate sample from one of the depths as a check on your precision.**
- **Remember to wear safety glasses and plastic gloves when performing these tests, both at home and in the boat.**

Specific Sampling Instructions:

Collecting and Fixing A Dissolved Oxygen Sample from the Water Surface

1. Before beginning the sampling, select one of the four depths (surface, 3 meters, 6 meters or near bottom) for performing the duplicate testing. The first month that you measure DO, choose any one of the depths for the duplicate. In each following month, rotate the depth at which the duplicate test is performed.
2. Remove the cap from the small clear water sampling bottle labeled **#A**. Reach into the lake and thoroughly rinse the bottle and cap **twice** with the water to be sampled. Be careful not to let the bottle slip from your hand.
3. Tightly cap the bottle and submerge it with both hands to a depth of about **6 inches**. Holding the bottle under water, remove the cap and allow the bottle to fill.
4. Tap the sides of the submerged bottle several times underwater to dislodge any air bubbles trapped inside. Replace the cap while the bottle is still submerged.
5. Pull the bottle out of the water. Examine it carefully to ensure that no air bubbles are trapped inside. If there are, repeat steps 3 and 4 to get an acceptable sample.
6. If you are duplicating the near-surface sample, repeat steps 2-5 with the sample bottle marked **“Dupe”**. Then, complete steps 7-13 on both bottles simultaneously.
7. Place the sampling bottle on a flat surface in your boat and remove the cap. Add **8 drops of Manganous Sulfate Solution** (the pink Chemical #1) to the sample. (Remember to hold the chemical reagent bottle vertically upside-down and squeeze **gently**.) Cap the chemical bottle before going to Step 8.
8. Add **8 drops of the Alkaline Potassium Iodide Solution** (clear Chemical #2) to the sample. Again, cap the chemical bottle.

Note: The Manganous Sulfate and Alkaline Potassium Iodide are added in excess. So, if you accidentally dispense 9 or 10 drops, it's OK. Do not start over. But you must put the Manganous Sulfate in first.

9. Cap the bottle labeled #A and mix by inverting gently several times. Set the bottle back down and allow the precipitate that has formed to settle below the shoulder of the bottle. This takes 4 to 5 minutes.

10. Invert the bottle several times again and allow the precipitate to settle below the shoulder again. This will take another 4 to 5 minutes.

11. Uncap the sample bottle and use the white measuring spoon to add **1 gram** of **Sulfamic Acid Powder** to the sample. This is **one level spoonful**. Cap the sample bottle and shake gently to mix. Continue mixing several minutes until the chemical and the precipitate have completely dissolved. The sample will now look clear yellow to brown-orange. (The Sulfamic Acid Powder is also added in excess. So, if you drop a few crystals, it is not critical. While adding the powder and other chemicals, be careful not to introduce air bubbles into the sample.)

12. Check to see if all of the precipitate is dissolved. Sometimes all of the grains of acid do not dissolve and bits of organic matter or sediment in the water may not dissolve. That's OK. Continue with the test.

13. The sample is now "fixed". Take the bottle back to your home to complete the rest of the steps of the titration. However, **do not wait more than 8 hours to complete the test**. You also no longer have to worry about adding air bubbles to the sample.

Collecting and Fixing Additional Samples

Using the LaMotte Sampler:

14. Place the glass sampling bottle marked #B in its holder inside the LaMotte sampler and close the sampler carefully as described in Sub-Task 4. Be sure the tubing is positioned inside the glass sample bottle. Then, lower the LaMotte sampler and retrieve a sample of water from **3 meters** deep. Check to be sure that all the bubbles escape from the sampler before pulling the sampler back into the boat. Keep the sampler vertical and check to be sure that the water level is above the top of the small dissolved oxygen bottle. Carefully remove the metal clip. Be sure the rope is out of the way so that the metal clip will open all the way out.

15. Reach in and grasp the dissolved oxygen bottle by the neck and lift it out of the sampler **slowly**. Place the bottle on a flat surface to perform the testing. Go back and repeat steps 7-13 to "fix" the dissolved oxygen sample.

16. Repeat steps 14 and 15 and 7-13 to collect and fix a sample from **6 meters** deep (#C). Also, repeat this process for **1 meter above the bottom** (#D) and for the **duplicate (Dupe)**.

Using the Vertical Sampler:

17. Use the vertical sampler to retrieve a sample of water from **3 meters** deep. Be sure that the white clip valve is closed on the discharge tube.

18. After retrieving the sampler, place the discharge tubing into the empty glass sampling bottle marked #B. Release the white clip and pull slightly on the upper ball (or open the

white stopper on the side of the sampler) to let in air and allow the water to flow out of the sampler. Rinse the bottle and the cap **twice** with the sample water and discard the water.

19. Insert the discharge tubing **all the way to the bottom** of the sampling bottle. **Slowly** release water to fill the bottle without introducing air into the sample. The end of the tubing should not rise above the level of the water in the bottle during filling. As the water begins to overflow the bottle, slowly withdraw the tubing while continuing to release water. When you are finished, the sample bottle should be completely full with no air bubbles. Tap the sides to be sure. If air bubbles are present, empty the bottle and try again.

20. Repeat steps 7-13 to add the chemical reagent and “fix” the sample. Take the fixed sampled back to your house to complete the titration.

21. To collect water samples from **6 meters** (bottle #C) repeat steps 17-19. Then repeat steps 7-13 to fix the samples. Repeat this process for the sample from **1 meter above the bottom** (#D) and **duplicate** (Dupe) samples.

Complete the Dissolved Oxygen Titration (back in your home)

22. Set the sample bottles and the chemical kit on a large flat surface. A kitchen counter is great. Work over the sink when dispensing chemicals. Do not contaminate the food preparation area. You may want to lay out a newspaper underneath to absorb any spills.

23. Remove the plastic top from the glass test tube. Pour **20 ml** of the fixed sample from sample bottle **#A** into the glass test tube. The amount of sample you pour into the test tube is critical. Make sure that the **bottom** of the meniscus is even with the white 20 ml line. (A meniscus is the concave curve that forms at the top of a liquid when it is in a tube).

24. Replace the plastic top on the test tube. Be careful not to tip over the test tube. If you do, there will still be enough fixed sample in the sample bottle to refill the test tube.

25. Fill the syringe with tap water to lubricate it. Expel the water from the syringe. Pump and shake the syringe to remove all the water droplets. Then, fill the syringe to the “0” mark with **Sodium Thiosulfate Solution** (the larger clear chemical #4). Do this by pushing the tip of the syringe into the Sodium Thiosulfate Solution bottle and inverting the bottle. **Slowly** withdraw the plunger until the tip of the black syringe plunger is even with the “0” mark. (If an air bubble gets into the syringe, just push the chemical back out until the bubble escapes. Then, continue filling the syringe.

26. Insert the filled syringe into the cap of the glass test tube containing 20 ml of the sample. **Add 1 drop** of Sodium Thiosulfate to the test tube. **Swirl** the test tube to mix the solution. Add another drop of Sodium Thiosulfate and swirl the tube. Continue this titration process **one drop at a time** until the color of the solution **just begins** to fade or get lighter. It will probably be a light yellow.

27. For the moment, set aside the syringe with Sodium Thiosulfate. **DO NOT empty the syringe.**

28. Add **8 drops** of **Starch Indicator Solution** (dark chemical #5) to the test tube. (Insert the dropper into the hole of the top of the test tube). Swirl the tube to mix. The solution

should turn from yellow to dark blue. (Exactly when and how much Starch Indicator to add is not critical, as long as the sample turns blue. The Starch Indicator serves to amplify the color change indicating when the dissolved oxygen has been completely used up. Use the Starch Indicator in the sink because it may stain).

29. Now, with the remaining Sodium Thiosulfate still in the syringe, continue the titration process. Add **one drop** at a time to the sample and swirl after each drop to mix the solution. Refill the syringe with Sodium Thiosulfate as described in step 25 if needed. Continue until the sample solution turns from **blue to clear**. Then **STOP**. Holding the test tube over a white sheet of paper is helpful in determining when the blue color disappears. (The amount of titration is critical. So, proceed slowly, swirling after each drop. The initial change to a clear solution is the stopping point. If the solution turns back to blue, ignore it).

30. Use the scale on the side of the syringe to determine the dissolved oxygen content of the water sample in milligrams per liter (mg/L). Read the number on the scale directly opposite the bottom of the black plunger tip. Each line on the scale equals 0.2 mg/L. If you had to refill the syringe for more Sodium Thiosulfate, add the number of mg/L that you read from the second syringe to the 10 mg/L that you used in the first syringe to get the total amount of dissolved oxygen. (As you will see, if the oxygen content is low, only a small amount of Sodium Thiosulfate is needed to complete the titration. If the oxygen content is high, more Sodium Thiosulfate is needed.)

31. Record the results in the appropriate blank on the data sheet.

32. Empty the contents of the test tube into a container with kitty litter. Do not flush into a septic system. Rinse the test tube with distilled water and dry. Empty the remainder of the Sodium Thiosulfate from the syringe into the kitty litter container. (If the container is large enough, keep it for several months testing. Then, seal the container, wrap in a plastic bag, and dispose in the regular garbage).

33. Repeat steps 22-32 on each of the remaining “fixed” samples, recording the results on the data sheet for 3 meters (#B), 6 meters (#C), 1 meter above the bottom (#D), and Dupe. Clean the sink and counter area, the test tube, and all of the sample bottles. Store the dissolved oxygen kit away.